

Amendments of the Claims:

1. (Previously presented): An isolated polynucleotide encoding a FEN-1 polypeptide as shown in SEQ ID NO:1 or SEQ ID NO:3, or a fragment of said polypeptide having flap endonucleolytic cleavage activity.
2. (Previously presented): An isolated polynucleotide, wherein said polynucleotide comprises a nucleotide sequence selected from the group consisting of SEQ ID NOS:29-51.
3. (Previously presented): An isolated polynucleotide of Claim 2, wherein said polynucleotide comprises the sequence of SEQ ID NO:28.
4. (Previously presented): A host cell comprising the polynucleotide of Claim 1.
5. (Previously presented): A non-mammalian host cell comprising a mammalian FEN-1 polypeptide of Claim 1.
6. (Previously presented): The polynucleotide of Claim 1 that is full length.

Claims 7-20 (Cancelled)

21. (Currently amended): A method of detecting the presence of a predetermined target nucleic acid sequence in a sample, comprising the steps of:

(a) contacting, under conditions in which a FEN-1 polypeptide exhibits cleavage activity, a sample suspected of containing a target nucleic acid comprising the predetermined target sequence of interest with:

(i) ~~a FEN-1 polypeptide;~~

(ii) (i) a 5'-polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region that is capable of specifically hybridizing under said cleavage conditions to a first portion of the target sequence nucleic acid and a 5'-region located immediately 5' to the 3'-region; and

(iii) (ii) a 3'-polynucleotide probe comprising a 5'-region that is capable of specifically hybridizing under said cleavage conditions to a second portion of the target sequence nucleic acid which is located immediately 3' to the first portion and a 3'-region located immediately 3' to the 5'-region,

~~under conditions in which~~ such that the 3'-region of the 5'-probe and the 5'-region of the 3'-probe specifically hybridize immediately contiguously with one another to the first and second portions, respectively, of the same target sequence nucleic acid molecule to form a 5',3'-double flap structure cleavable by a FEN-1 polypeptide; and

(b) cleaving the 5',3'-double flap structure with a FEN-1 polypeptide; and

~~(b)~~ (c) detecting the presence or absence of, and/or quantifying the amount of, FEN-1 polypeptide-generated cleavage, thereby detecting the presence of the target sequence nucleic acid in the sample.

22. (Previously presented): The method of Claim 21 in which the 5'-probe contains a detectable label.

23. (Currently amended): The method of Claim 22 in which the 5'-region of the 5'-probe comprises a 5'-flap region that contains a the detectable label.

24. (Currently amended): The method of Claim 23 in which the 5'-end of the 5'-probe contains a the detectable label.

25. (Previously presented): The method of Claim 21 in which the 5'-probe is immobilized on a support.

D 26. (Currently amended): The method of Claim 21 in which the FEN-1 polypeptide is ~~a mammalian or a yeast FEN-1 polypeptide~~ encoded by a polynucleotide comprising a sequence selected from the group of sequences consisting of SEQ ID NOS: 29-51.

27. (Currently amended): The method of Claim 26 in which the FEN-1 polypeptide is ~~a yeast FEN-1 polypeptide, a murine FEN-1 polypeptide or a human FEN-1 polypeptide~~ encoded by a polynucleotide comprising SEQ ID NO:28.

28. (Currently amended): The method of Claim 26 21 in which the FEN-1 polypeptide is ~~a human FEN-1 polypeptide comprising~~ comprises the amino acid sequence shown in SEQ ID NO:1 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

29. (Currently amended): The method of Claim 26 21 in which the FEN-1 polypeptide is ~~a murine FEN-1 polypeptide comprising~~ comprises the amino acid sequence shown in SEQ ID NO:3 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

30. (Currently amended): The method of Claim ~~26~~ 21 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide comprising comprises the amino acid sequence shown in SEQ ID NO:5 or SEQ ID NO:7 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

31. (Currently amended): The method of Claim 21 in which the 3'-region of the 3'-probe ~~comprises a 3'-flap region that~~ is 1 to 10 nucleotides in length.

32. (Currently amended): The method of Claim 21 in which the ~~3'-flap region~~ 3'-region of the 3'-probe is 1 nucleotide in length.

33. (Currently amended): The method of Claim 21 in which the 5'-region of the 5'-probe ~~comprises a 5'-flap region that~~ is 1 to 20 nucleotides in length.

34. (Previously presented): The method of any one of Claims 21-33 in which the amount of FEN-1 polypeptide-generated cleavage is quantified.

35. (Previously presented): The method of any one of Claims 21-33 in which the presence or absence of FEN-1 polypeptide-generated cleavage is detected.

Claims 36-50 (Cancelled)

51. (Previously amended): A hybridization complex comprising:
(a) a bridge polynucleotide comprising a first portion and second portion located immediately 3' to the first portion;
(b) ^{of SEQ ID NO: 1 or 2} a first polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region and a 5'-region located immediately 5' to the 3'-region; and
(c) a second polynucleotide probe comprising a 5'-region and a 3'-region located immediately 3' to the 5'-region,
wherein the 3'-region of the first probe and the 5'-region of the second probe are specifically hybridized immediately contiguously with one another to the first and second portions, respectively, of the same bridge polynucleotide molecule, thereby forming a hybridization complex.

52. (Previously presented): The hybridization complex of Claim 51 in which the first probe contains a detectable label.

53. (Currently amended): The hybridization complex of Claim 52 in which the 5'-region of the first probe contains a the detectable label.

54. (Currently amended): The hybridization complex of Claim 53 in which the 5'-end of the first probe contains a the detectable label.

55. (Previously presented): The hybridization complex of Claim 51 in which the first probe is immobilized on a substrate.

56. (Currently amended): The hybridization complex of Claim 51 in which the 3'-region of the second probe ~~comprises a 3'-flap region~~ that is 1 to 10 nucleotides in length.

57. (Currently amended): The hybridization complex of Claim 56 in which the ~~3'-flap region~~ 3'-region of the second probe is 1 nucleotide in length.

58. (Currently amended): The hybridization complex of Claim 51 in which the 5'-region of the first probe ~~comprises a 5'-flap region~~ that is 1 to 20 nucleotides in length.

59. (Currently amended): A kit for use in detecting the presence of a predetermined target nucleic acid sequence in a sample, comprising:

- (a) a FEN-1 polypeptide;
- (b) a first polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region capable of specifically hybridizing under FEN-1 polypeptide cleavage conditions to a first portion of a the predetermined target sequence ~~nucleic acid of interest~~ and a 5'-region located immediately 5' to the 3'-region; and
- (c) a second polynucleotide probe comprising a 5'-region capable of specifically hybridizing under FEN-1 polypeptide cleavage conditions to a second portion of the target ~~nucleic acid~~ sequence which is located immediately 3' to the first portion and a 3'-region located immediately 3' to the 5'-region,

wherein the 3'-region of the first probe and the 5'-region of the second probe are capable of specifically hybridizing immediately contiguously with one another to the first and second portions, respectively, of the ~~same target nucleic acid molecule~~ sequence to form a 5',3'-double flap structure that is capable of being ~~bound or~~ cleaved by the FEN-1 polypeptide.

60. (Previously presented): The kit of Claim 59 further in which the first or second probe contains a detectable label.

61. (Previously presented): The kit of Claim 59 in which the FEN-1 polypeptide contains a detectable label.

62. (Currently amended): The kit of Claim 59 in which the 3'-region of the second probe comprises a 3'-flap region that is 1 to 10 nucleotides in length.

63. (Currently amended): The kit of Claim 59 in which the 3'-flap region 3'-region of the second probe is 1 nucleotide in length.

64. (Currently amended): The kit of Claim 59 in which the 5'-region of the first probe comprises a 5'-flap region that is 1 to 20 nucleotides in length.

65. (Previously presented): The kit of Claim 59 in which the first probe contains a detectable label.

66. (Currently amended): The kit of Claim 59 65 in which the 5'-region of the first probe comprises a 5'-flap region that contains a the detectable label.

67. (Currently amended): The kit of Claim 66 in which the 5'-end of the first probe contains a the detectable label.

68. (Previously presented): The kit of Claim 59 in which the first or second probe is immobilized on a substrate.

69. (Currently amended): The kit of any one of Claim 59-68 in which the FEN-1 polypeptide is a mammalian or a yeast FEN-1 polypeptide encoded by a polynucleotide comprising a sequence selected from the group of sequences consisting of SEQ ID NOS: 29-51.

70. (Currently amended): The kit of ~~Claim 69~~ any one of Claims 59-68 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide, a murine FEN-1 polypeptide or a human FEN-1 polypeptide encoded by a polynucleotide comprising SEQ ID NO. 28.

71. (Currently amended): The kit of ~~Claim 70~~ any one of Claims 59-68 in which the FEN-1 polypeptide is a human FEN-1 polypeptide comprising comprises the amino acid sequence shown in SEQ ID NO:1 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

72. (Currently amended): The kit of ~~Claim 70~~ any one of Claims 59-68 in which the FEN-1 polypeptide is a ~~murine FEN-1 polypeptide comprising~~ comprises the amino acid sequence shown in SEQ ID NO:3 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

D 73. (Currently amended): The kit of ~~Claim 70~~ any one of Claims 59-68 in which the FEN-1 polypeptide is a ~~yeast FEN-1 polypeptide comprising~~ comprises the amino acid sequence shown in SEQ ID NO:5 or SEQ ID NO:7 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.
